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GOLDIC® – Next Generation Regeneration

CHAPTER

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GOLDIC® is the next generation of stem cell therapy with the help of specially designed gold particles using patients own blood. GOLDIC® stands for “GOLD INDUCED CYTOKINES”. Cytokines are proteins that regulate the growth and differentiation of cells. Cytokines are a group of peptides, which mainly initiate or regulate the growth, the proliferation and differentiation of target cells. Cytokines are steadily gaining importance in medicine and in cellular biology.

GOLDIC® – PROCEDURE

In this procedure a small quantity of the patient’s blood is taken and then it is prepared through a process with the help of specially designed gold particles. The shape, number, surface, concentration and size of the gold particles play a decisive role here. The regenerative factors (cytokines and various growth factors) from the patient’s own body are filtered and concentrated. This optimized serum produced naturally in the body is then injected in the painful areas without other additives. The objective is to stimulate the body’s own power in such a way that damaged or injured tissue can be regenerated. This is done with the help of stem cells from the body itself, which are activated and differentiated.

The key protein in case of GOLDIC® - procedure is Gelsolin. The Gelsolin level of patients in plasma (pGSN) in conjunction with other cytokines is especially important for successful treatment. pGSN is the longest known member of a family of actin- binding proteins. pGSN is an important protective protein and it prevents the toxic reaction, which occurs when cells die or go through apoptosis. Moreover, pGSN causes the differentiation of body’s own cells, which are necessary for regeneration of tissues. It is known in literature that in all types of injuries, inflammations or degenerative processes can lead to a significant reduction of pGSN (Suhler et al 2004).

Furthermore, important stem cell specific factors like G-CSF (granulocyte-colony stimulating factor) and SCGF-IS [hematopoietic - stem cell growth factor - beta) are produced to a significantly high degree using the GOLDIC® - procedure. Through the contact of the patient’s blood with the gold particles, the white blood cells (leukocytes) and mainly the monocytes are activated. Amongst the white blood cells the monocytes are in the best position to neutralize and phagocytize, it literally means to “devour”, bacteria and tissue debris. These two proteins can activate the stem cells in the bone marrow and will be released to the systemic circulation system (Schneider, et al. 2017).

GOLDIC® can be seen as a novel approach using activated serum that stimulate medicinal signaling cells on sites of injury or disease. The secretion of bioactive factors can act in an immunomodulatory and regenerative way.

Detection of the Chondroprotective Effect of GOLDIC on the In-Vitro Cartilage Droptower Model

The influence of mechanical overload on the articular cartilage in the context of osteoarthritis development is sufficiently known. Well-established and validated in vitro models are available to investigate the influence of mechanic stress in cartilage bone systems (Huser, et al. 2006). A good indicator of the beginning cartilage degeneration is the measurement of the proteoglycan content in the supernatant.

In an animal-experimental study in the Göttingen Minipig, 4 syringes were drawn and processed in 6 animals according to the GOLDIC® production protocol. The necessary cartilage-bone samples of the respective animals were obtained under aseptic conditions from the respective hip heads and cut into 8x8x10 mm (length/width/height) blocks. The cartilage was macroscopically intact in all ex-planning. Further storage until impact was carried out in DMEM medium with the addition of 10% bovine serum, 100 U/ml penicillin, 100 micrograms/ml gentamycin and 1.25 U/ml amphotericin B.

The impact test was carried out on the day of the sampling. An axial impact was used with a cylindrical “drop tower” of polymethyl methacrylate, which had a height of 33 cm and a diameter of 4 cm. The cylindrical weight was 493 g and had the dimensions of 5 cm in height and 3.95 cm in diameter. This was connected to the one separate round impactor piece (weight: 7 g, height: 1 cm, diameter: 0.6 cm). The impact was made once per explant by the free fall of the impactor in the “drop tower” from a height of 15 cm under sterile conditions. The impact load was 0.736 J on a cartilage surface of 28.3 mm².

The explants were randomized in 3 groups:

- ☐ Non-impacted controls
- ☐ Impacted untreated controls
- ☐ Impacted preparations with GOLDIC additive (300 µl) on day 0 and 7

After the impact procedure, the bone cartilage preparations were washed 3 times with PBS and transferred to 12-well culture plates. Each explant was planted with 3 ml of the respective treatment medium and incubated under standardized conditions (37°C, 5 % CO₂) in the incubator. The media was changed every 72 hours. The conditioned GOLDIC serum was added to the medium on day 0 and 7. The proteoglycan content in the supernatant was measured on day 2, 7 and 14. For this purpose, the Blyscan Glycosaminoglycan Assay of Biocolor Ltd., (Carrickfergus, UK) was used. The mean GAG concentration was given in µg/ml medium.

The analysis of PG release shows in the group of impacted preparations a significant mockery of the concentration of PG over time. Treatment with GOLDIC significantly reduced PG release. This reduction was comparable to the values of a non-impacted control group.

The treatment of impacted cartilage-bone explants with a with the GOLDIC process thus has a detectable chondroprotective effect.

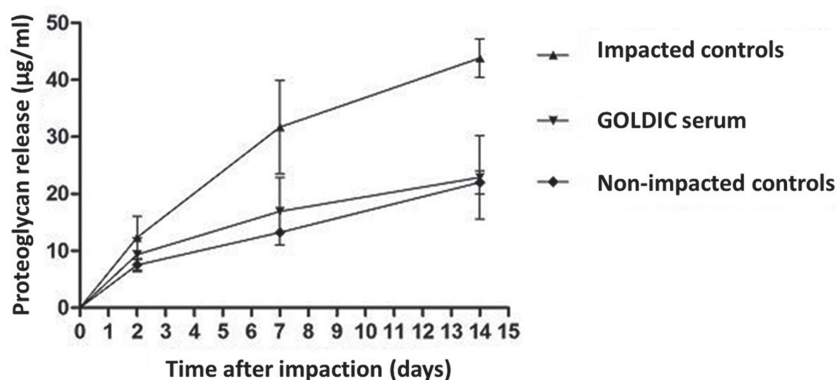


FIGURE 47.1 Results of an in-vitro study using osteochondral explants treated with drop tower impaction. The documentation of proteoglycan (PG) concentration in the supernatant demonstrated a significant decrease of PG release when Goldic serum is added to the media.

Clinical Outcome of GOLDIC® – Procedure in Knee Osteoarthritis (KOA)

A prospective case study was conducted to evaluate the effects of intra-articular GOLDIC® therapy in patients with osteoarthritis of knee joints. All the patients were recruited for the study in the 2013 and were followed for a period of 60 months after the last GOLDIC® injection. All the patients gave written informed consent for treatment with 4 intra-articular GOLDIC® injections for treatment of osteoarthritis of knee joint. The clinical use of gold-induced autologous serum (Goldic®) was reported to the competent regional council in accordance with § 13 Abs.2b AMG and study was conducted according to the guidelines of the good clinical practice (GCP) guidelines.

Patients

Patients with osteoarthritis of knee joint with minimum 4 months of knee pain and/or swelling and radiological evidence of joint degeneration on x-rays or MRI were included in this study. Patients with joint infection, pregnancy, neurological disorder, spondyloarthropathies, gout, hyperlipidemia, rheumatoid arthritis, sarcoidosis, any metal sensitivity or pathologies of the lower limb which would interfere with the evaluation of KOA were excluded from the study. Patients were also excluded if they had received intra-articular injections, symptomatic slow-acting drugs in osteoarthritis (SYSADOA), oral or topical steroids and/or non-steroidal anti-inflammatories (NSAIDs) in the last two months.

Twenty-eight patients (31 knees) having osteoarthritis of knee joints who fulfilled the inclusion criteria were enrolled in this study. These patients were treated with 4 intra-articular GOLDIC® injections. All the patients were evaluated by visual analog scale (VAS) before injection and every 12 months up to 5 years after the injections. All the adverse events and complications were recorded during the follow-up period.

GOLDIC® PROCEDURE

For GOLDIC® procedure, 4 × 10 mL of blood is collected from each patient using four GOLDIC® BTS syringes (Arthrogen GmbH, Gmund, Germany) and is incubated at 37°C for 24 hours. After the

incubation process, the blood is centrifuged at 4000 rpm (2250 g) for 10 minutes. Then supernatant conditioned serum is collected and filtered through a 0.22 μ M syringe tip filter (Millex GP, Merck Millipore, Tullagreen, Carrigtwohill, Cork, Ireland); and finally used for intra-articular injection or can be stored at -20°C later use. Patients received 4 intra-articular injections of 3 ml GOLDIC® serum every 3-6 days. All the patients received ultrasound guided intra-articular injection by standard lateral approach under all aseptic precautions with 22G needle. Patient were advised to use ice pack/cold therapy for any knee pain at home and to avoid long walking/standing for next 24 hours. Patients were instructed to use only Paracetamol (1 g) for post-injection pain and to strictly avoid NSAIDs. During the treatment period patients were asked to avoid strenuous exercises, and were allowed only non-impact exercises like cycling and pool exercises; and subsequently the gradual resumption of normal sport or recreational activities was allowed.

RESULTS

A total of 28 patients (31 knees) having osteoarthritis of knee joint were included in the study. There were 13 male and 15 female patients; mean age was 57.7 years (range, 45-81 years); 18 patients had right knee and 13 had left knee involvement. All knees had stage 3 or 4 in Kellgren and Lawrence score.

All 31 knees showed statistically significant improvement in the mean of VAS score at all timepoints compared to baseline ($P < 0.001$). Two patients failed treatment, and underwent total knee replacement after 2 years, no patients were lost to follow-up. Only minor adverse events were detected, such as a mild pain and effusion after the injections, which persisted for not more than 2 days in few patients.

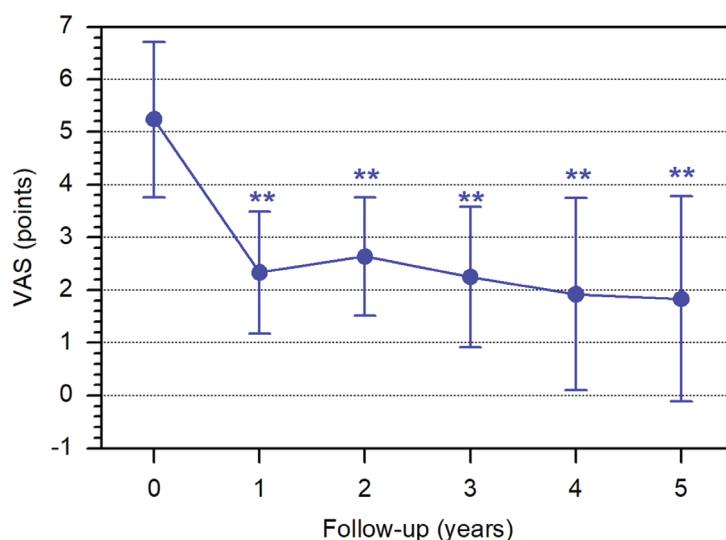


FIGURE 47.2 VAS-scores for baseline, 12, 24, 36, 48 and 60 months after treatment with GOLDIC injections. A significant improvement of pain could be seen at all time points compared to baseline (** $p < 0.001$).

GOLDIC® - PROCEDURE IN COMPARISON WITH OTHER ORTHOBIOLOGICAL PROCEDURES

Stem Cell-based Treatments

Stem cells that are collected from the bone marrow or from the adipose fat tissue (operation under general anesthesia with all possible risks and side effects), must still be processed to obtain an adequate number of stem cells. Injecting these stem cells is possible only after a few weeks due to the time required for the cultivation and differentiation. In addition, adult stem cells are not practicable for all body tissues and are difficult to obtain. Restricted growth and conditional transformability limit their use. The cost factor will also play a significant role here. Even stem cells age and thus old stem cells are less functional than younger ones. Theoretically, it must be considered that if the cultivation is done with the same stem cells several times, there can be a risk of degeneration of individual stem cells. Present research shows that the stem cells stimulate medicinal signaling cells on sites of injury or disease. The transplanted cells are not involved in trophic mechanisms, instead the local cells secrete the bioactive factors that will initiate the healing process (Caplan 2017).

PRP and ACP (Autologous Conditioned Plasma) Procedure

The normal PRP (platelet-rich plasma) procedures mainly use the thrombocytes obtained from the blood plasma of the patient as well as growth factors and at most a relatively low number of stem cells. In this way the inflammation in the damaged tissue / organ / joint can be reduced, the important gelsolin level is decreased in this process and not increased as in case of the GOLDIC® -procedure. PRP and ACP does not seem to promote long lasting regeneration or restoration of joint homeostasis as multiple injections over a 9-month period is required. The literature showing results of PRP is debatable and conflicting; studies reports PRP as better (Xie, et al. 2014), having same effect (Cole, et al. 2015) or worse (Filardo, et al. 2017) than hyaluronic acid in relieving pain of osteoarthritis of knee. Duration of effect of PRP has also been shown to be variable lasting for 6 months to one year (Holgiun 2014). The variable results could be explained by a lack of standardized technique of PRP preparation; variable types (Gholami, et al. 2016, Mlynarek, et al. 2016, Filardo, et al. 2015), or other factors like gamma interferon (Braun et al 2014), the number and site of PRP injections. In comparison to PRP, the technique of GOLDIC® preparation is standardized and uniform, so content and quality of the GOLDIC® serum will always will be the same and it will not affect the outcome of treatment.

Although, in-vivo studies demonstrate that ACS indeed improves pain and function in humans having osteoarthritis of joints (Gholami, et al. 2016), however one in-vitro study showed that ACS has increased levels of both anti-inflammatory and pro-inflammatory cytokines (tumor necrosis factor- α) and when it was applied to a osteoarthritis explant tissue, it did not have any direct effect on cartilage metabolism (Braun, et al. 2014)

The GOLDIC® - procedure is a platform technology in which all kind of damaged connective tissue can be treated. It is well suited for treating acute and chronic orthopedic diseases. Compared to other orthobiological procedures the GOLDIC® technology has an excellent long-term efficacy. The reason for this can only be explained by the high regenerative potential of this procedure.

The exact mode of action of the GOLDIC® procedure is not well understood. In-vitro studies have shown a significant increase in plasma gelsolin level in the autologous serum and increased gelsolin level in synovial fluid after every GOLDIC® injection. Both gelsolin and G-CSF have been shown to promote regeneration. Gelsolin is an actin-binding protein and occurs in both cellular cytoskeleton and in the plasma (Silacci, et al. 2004). The cytoskeleton is responsible for the viscoelasticity of cells (Trickey, et al. 2004). Gelsolin also regulates other important cell functions including cell motility, phagocytosis, apoptosis and the activation of thrombocytes (Silacci 2004), and its plasma concentration is decreased in various tissue degenerative diseases (Suhler, et al. 1997). Experiments have shown decreased plasma gelsolin concentrations in animals with sepsis, and treatment with gelsolin had a positive effect on the survival rate in these animals (Lee, et al. 2007). Plasma gelsolin serves as a buffer to intercept inflammatory reactions of the body (DiNubile, et al. 2008) and was found to be decreased in rheumatic arthritis (Osborn, et al. 2008). Interestingly, the gelsolin level were found more reduced in the affected joints in comparison to plasma level. Therefore, it is an important finding that the intra-articular treatment with gold-activated serum (GOLDIC®) resulted in a significant increase of gelsolin in the synovial fluid (Schneider, et al. 2019).

CONCLUSION

GOLDIC® - procedure is a promising new approach to treat degenerative and inflammatory disease. The upregulation of gelsolin and presence of other growth factors in GOLDIC® appears to be contributing in restoration of overall biological homeostasis. Despite the promising early clinical results, randomized controlled studies with long term follow-up are required to validate the real potential of GOLDIC® and to compare these good results to other blood-based platforms.

RECOMMENDED LITERATURE

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